

BULLETIN No. 9

Vol. II

□ MARCH, 1923 □



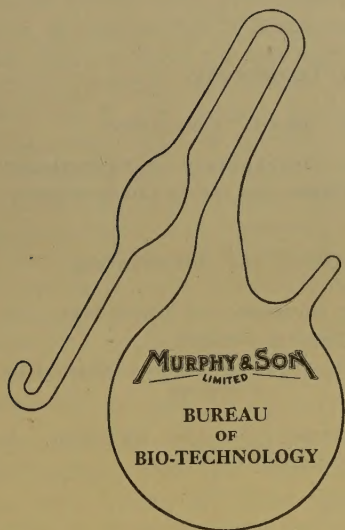
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The "Biotec" Comparator

THE interest already shown in our new "Biotec" Comparator leads us to assume that a description of the instrument together with details of our own mode of employment may be appropriately included here. The modern conception of acidity, presenting a new outlook to biological science, has compelled laboratory workers to provide themselves with some means of ascertaining the hydrogen ion concentration of the fluids with which they have to deal. This has meant in most instances the improvising of an illuminated comparator block into which test-tubes containing fluids plus indicator might be placed, and their colours compared by transmitted light. The illumination of the block is, of course, the important point, and constitutes the chief difficulty. Innumerable alterations and adjustments to our first improvisation evolved the apparatus as used at present in our laboratories, and now offered for sale.

Primarily the blocks themselves demanded attention, and we came to the conclusion that they must be constructed of hard, close-grained wood, finished with some material resistant to the chemicals liable to be spilled over the surfaces. The interior surfaces are dull black. The distance between the front and rear of the blocks, considered in relation to the size of the test-tubes employed, is of paramount importance in obtaining bright evenly illuminated patches of colour. Ground glass is essential at the front of the block, and its use in rear also is recommended.

Two blocks are required: one for colourimetric determination of H.I.C., and the other for the titration of biological fluids which, on account of their original colour, cannot be titrated in the ordinary way, or which require to be titrated to a particular H.I.C. It has hitherto been the practice to mount these two blocks

separately, or to have some means of interchanging them upon one fixture. In either case difficulties are unnecessarily involved, and we have mounted both blocks permanently upon the same fixture in such a way that they are available without further adjustment.

Turning to the question of illumination, it is obvious that we require to receive a strongly and evenly lighted area upon the back of the block emerging from a point in alignment with the observation holes. The enclosure of an "opal" electric bulb in a tinned iron case of suitable dimensions meets this demand adequately without resorting to optical devices. Both blocks are

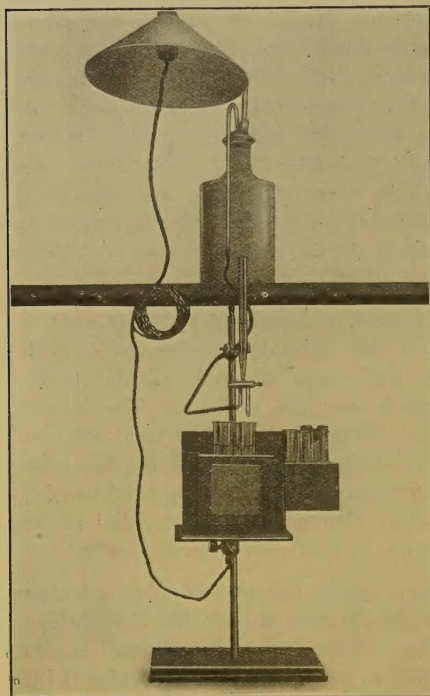


FIG. 1.

affixed to the metal body in positions of alignment with the luminant. The generation of heat within the instrument is unavoidable, but ventilation holes are bored at suitable points and a switch is conveniently placed under the base in order that the current need not be turned on except during a determination. Heating of the apparatus itself, with a consequential error in determinations, cannot occur unless the current is allowed to remain switched on for periods far exceeding those required for any determination.

The base of the apparatus is constructed of 5-ply board and shows no tendency to warp on account of the heat.

A removable slide in the metal body renders the luminant readily available for microscopic or tintometer lighting.

For the determination of pH by the colourimetric method we have found the colour standards of Medalia especially convenient and of sufficient accuracy for routine work. Similar colour standards evolved by Gillespie, or Barnet and Chapman, may be employed. Medalia's colour standards were described in *Bulletin* No. 3, and whilst it is admitted that they cannot be considered adequate substitutes for their corresponding buffer mixtures, a considerable amount of reliable work may be done with them. Our comparator block is bored to take the nine tubes used in this method, but this does not in any way interfere with the use of only six tubes when buffer mixtures are employed as standards.

In order to facilitate the titration of fluids by means of the comparator, we have effected one or two improvements of existing methods. The block is bored to take four 1 in. boiling tubes (these conveniently hold 50 cc.) Tubes Nos. 1 and 2 are in front of Nos. 3 and 4 respectively. When it is required to titrate to any particular pH, No. 3 tube contains the 50 cc. of the appropriate buffer mixture plus indicator. Sodium acetate will provide pH 7 when neutrality is to be the end point.

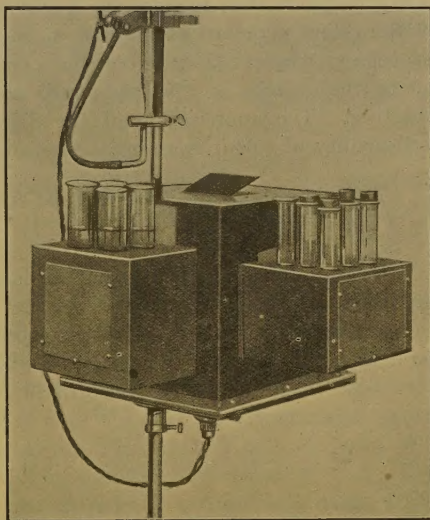


FIG. 2.

Tube No. 4 contains water alone. Nos. 1 and 2 contain 50 cc. of the fluid to be titrated, and to No. 2 is added the same amount of indicator as in No. 3.

The complete comparator is designed to fit upon the usual 24 in. or 30 in. by $\frac{1}{2}$ in. type of iron retort stand. The upright passes through the instrument, which may then revolve in a horizontal plane. By this means either block may be brought to the front for use. Above the instrument is placed a clamp carrying the burette used in the titration and this is adjusted to deliver into tube No. 2A. A slight rotary movement to the right will bring No. 1 under the burette so that the necessary amount of fluid may be added to that tube to compensate for dilution of colour in No. 2. In many cases this compensation for dilution may be carried out by means of water from another burette; but many coloured biological fluids change their colour slightly during titration, and compensation must be made for this. It will be noticed that No. 1 tube (fluid alone), and not the colour standard, in this case, is adjusted, although the titration tube (No. 2), also containing indicator, receives the addition of solution. A dilution of the colour standard would only mean an alteration in *intensity* of colour, whereas dilution of the compensation tube actually alters the *quality* of the composite colour from both viewed together. The appreciation of the difference between intensity and quality of colour is essential when using the comparator.

The contents of the tube may be stirred during the titration by a glass propeller.

In assembling this instrument we have adhered to the use of ordinary laboratory apparatus as far as possible, and the component parts are as simple as is practicable. Our aim has been to produce this commercial and efficient type of instrument at a cost which might be met without difficulty in the routine works' laboratory.

P. H.

The Decay of Structural Timber

ITS CAUSES AND PREVENTION

FROM the nature and frequency of enquiries received either directly or through our representatives who move about among users of timber in every department of industry, it is evident that a good deal of misconception exists in regard to the causes of the deterioration and destruction of wood. The subject is one of the gravest importance to the owner of property, whose capital is largely represented by buildings and premises composed more or less of timber, yet, frequently, he remains quite unconscious of the fact that the items "depreciation" and "repairs," in his annual account of profit and loss may be credited to the baneful action of living organisms.

The decay of timber from one cause or another is in constant progress. From the moment a tree is felled, and indeed often long before that time, agencies are at work which, left to continue their effects unchecked, speedily reduce the soundest oak, pine or other kind of forest timber to a condition of rottenness, and finally to humus which goes to enrich the soil from which it sprang. That is one of the processes whereby the balance of nature is maintained, and except under climatic or physiographical conditions inhibitive to biogenic action—such as inundation by water—it requires the intervention of man to prevent it. More precisely, the term "decay" has reference to the gradual change, both chemical and physical, which wood undergoes in its conversion from a compact, cellular structure of complex chemical constitution into a greatly reduced mass of structureless substances, of relatively simple composition, by the action of living organisms.

Insects are common agents of destruction and are frequently contributory factors in hastening decay by other means. The seriousness of their ravages has recently received emphasis in the newspaper reports of the costly work that has been incurred in

renovating the roofing timbers of St. Stephen's Hall, Westminster. Almost irreparable damage had resulted from the slow but continuous tunnelling operations of one of the well-known death-watch beetles. Wood used in cabinet and furniture making suffers considerable damage from the boring action of beetles known as "worms," and expensive woods like mahogany and teak may become so perfectly riddled as to crumble to powder. Bacteria play some part in the destruction of wood, more particularly in the final stages of decay, but the damage is negligible compared with that wrought by the higher fungi, which are the general agents of decay in this, as in all temperate countries.

Among some half-dozen species of fungi which most constantly attack and destroy timber used in the construction of buildings, three are responsible for the greater bulk of the damage sustained in this country. What might be called the diseases of wood due to these fungi are mostly referred to as "dry-rot," although the name "dry-rot fungus" is usually applied to one of them, *Merulius lacrymans*. The two other species are *Lenzites saepiararia*, occurring more particularly on coniferous woods, and *Coniophora cerebella*,¹ a species less impartial in its selective habit, and in the writer's experience, growing more frequently on worked timber out of doors than inside buildings.

Although the three foregoing species have been mentioned as being the chief offenders it by no means follows that a serious case of destruction must be due to one of them. Not infrequently the ravages of some little-known species are brought to light and a fungus unsuspected of economic interest, after investigation, may prove to be of the greatest importance from this point of view. A recent case in point is that mentioned in a report of the French mycologists, Mangin and Patouillard,² "On the rotting of the timber-work in the palace of Versailles by *Phellinus cryptarum* Karst."³ So far as

¹ This fungus, like many other common species, is burdened with a number of names, and whilst this is the one used in an extensive literature on the destruction of wood by fungi, it is described in modern text books as *Coniophora puteana*.

² Comptes Rendus Acad. des Sciences, cxxv, 9, 1922, through Rev. App. Mycology, 2, 1923, p. 97.

³ Known in this country as *Polyporus cryptarum* and found only on rotting coniferous wood.

those authors are aware the fungus has never been previously recorded as destroying woodwork in buildings, but they find that it plays the chief part in decay of the massive oak beams in the roof of the Louis XIII. wing of the palace named above, as well as in another case of the rotting of oak beams supporting a ceiling, which occurred in a room kept closed for fifteen years in a school at Etampes. With respect to the palace timbers, the authors state that good conditions for its development are supplied by the fact that the oak beams have been covered with plaster having an outer air-tight coating which prevents air and light reaching the wood. The attacked timbers, especially the ends embedded in the masonry, are reduced almost to the consistency of lint, having long fibrous strands extending lengthwise in the beams. The decayed wood is easily crushed together by slight pressure, but does not crumble to powder as is the case with wood destroyed by the dry-rot fungus, *Merulius lacrymans*. A full investigation of this fungus leads Mangin and Patouillard to the conclusion that it is a very dangerous destroyer of wood.

Dry-rot is undoubtedly the greatest cause of trouble in this country. Here it attacks floor-boards, joists, skirting-boards, shelving, wooden roofs and their supporting timbers in houses, mills, warehouses, breweries, etc., as well as the wooden vessels, vats and other equipment used by tanners, dyers, distillers, vinegar makers, chemical manufacturers and others. Fig. 6 is a photograph taken in one of the largest buildings in the North of England. It shows one end of a beam attacked by dry-rot. Without having made much lateral progress from its starting point in the end embedded in a brick wall, the timber has become so much rotted as to have sunk nearly the depth of one brick and to necessitate support by an upright, which, by the way, is itself supported by a floor attacked by dry-rot, and from which the beam-attacking fungus probably originated. Only a corridor is shown in the photograph, but when it is explained that the beam is continued through the partition to the right and forms one of the supports to a roof of 60 ft. span, it will be gathered that sooner or later there will be a costly job which is here seen in the making. There is hardly any kind of wood which, at normal temperatures, this fungus will not attack, and all authorities are agreed that the

annual damage caused by its action on structural timber must run into millions of pounds sterling.

Some insight into the nature and destructive action of the dry-rot fungus, and similar organisms, leading up to a discussion of methods of prevention will be more readily obtained by a brief consideration of the anatomical characteristics of the wood which it attacks.

The trunk of a forest tree, from which a plank or beam is sawn, may be likened to a geometric cone with the apex pointing upwards, although, for all practical purposes, a portion of the trunk between any two cross sections may be regarded as a cylinder. The structure of such a cylinder is essentially the same whether its diameter be a few inches or several feet, and the examination of three sections, a transverse, radial and tangential section, respectively, will reveal the more important features of its structure. The diagram Fig. 3 makes it clear what aspects are shown by these three sections. Coniferous wood, represented commonly in this country by deal, differs in certain details readily seen microscopically, but from the point of view of fungus attack its structure makes no material difference. It will be seen in the photograph of a tangential section of a deciduous wood (Lime) Fig. 4, that the wood consists primarily of a series of tubes. These fit into each other at either end and form bundles of columns running parallel to the long axis of the stem. The fact that they are actually tubes is shown by the transverse section, Fig. 5, which cuts across them at right angles. It will be seen at A

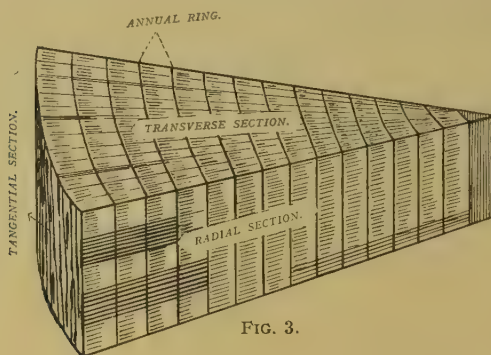


FIG. 3.

and B in the cross section of an annual ring that these tubes are of two general diameters, larger tubes nearer the pith (the central portion) and an outer row of tubes of smaller diameter. These represent the production of tissue at two different periods in the growth of the tree, the larger tubes being formed in the spring when the activity of the tree is at its greatest, the smaller ones during the summer; the two zones together make up what is known as the annual ring. The only material way in which the appearance of a section of a large tree would deviate from that of a smaller tree or branch would be by the greater number of annual rings, according to the age of tree from which the section had been taken.



FIG. 4.

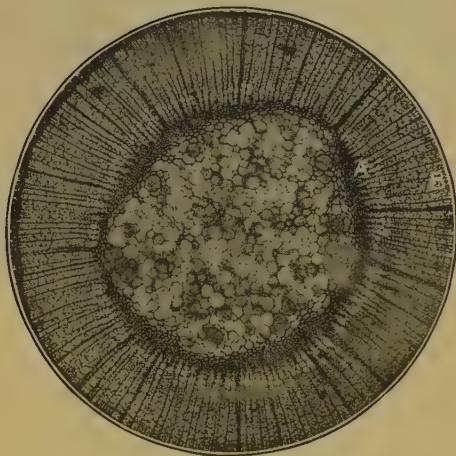


FIG. 5.

The tubes to which reference has been made are not so simple as might appear from the foregoing remarks ; many kinds of tissue and vessels enter into their more minute construction, and, moreover, the system of arrangement is complicated by the existence of plates of cells running at right angles to the tubes, from the central pith outward toward the bark. The latter are the medullary or pith rays, which maintain a connection between the central and the outer portions of the stem for the purpose of conveying water and food substances. It is these rays which give the silver grain to oak, beech and other woods, and an end view of the cells composing them is well shown in the photomicrograph, Fig. 4, and diagrammatically in the radial and tangential sections, Fig. 3.

The structure of the bark and other portions of the outer layers of the trunk need not be considered, as they are always removed in the process of dressing the timber for market. It is important to note, however, that the newer wood nearer the bark, known as the sap wood, in distinction from that of the older portions of the trunk called the heart wood, always contains and persistently retains a greater percentage of water than does the heart wood ; on that account and also because it contains readily assimilable sugars it is more susceptible to the attacks of fungi.

What has been said is sufficient to indicate the character of wood structure and to show that there is a system of tubes and cells, each of which, it is to be understood, is capable of accommodating an organism or the portion of an organism smaller than itself and possessed of some means of reaching it. This, indeed, is exactly what happens in an attack by a fungus, and small as the diameter of the wood vessel is, the fungus thread is smaller, and it enters the cell or tube through a hole which it bores for the purpose. A comparison of the size of the fungus thread and the tube may be obtained by taking a 2 in. wire nail and comparing it with an ordinary fountain pen.

A fungus, whether it be *M. lacrymans* or any other species, consists primarily of a vast number of fine colourless threads, each of which is called a hypha. These hyphæ together—and there may be few or many millions of threads—form the mycelium or vegetative portion of the fungus, which is always the part



FIG. 6.

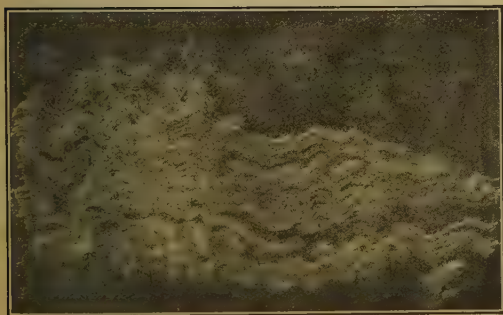


FIG. 9.

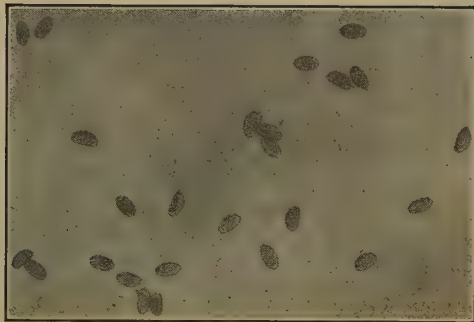


FIG. 8.



FIG. 7.

directly responsible for any damage done either to a living plant or to dead wood. Sooner or later, after spreading throughout its host, the mycelium will form large masses of compact material, on which some of the hyphæ develop specialised cells for the purpose of bearing reproductive bodies. This is the fruiting stage and the reproductive bodies are spores. In Fig. 7 the fruiting layer of *Merulius lacrymans* is shown where it has developed on a deal board that had been used in comparatively new repairs to a roof. It is seen to form rust-coloured, cushion-like masses, with a reticulated surface and white margin; the reticulations constitute the edges of pores, which are lined with a delicate membrane covered with basidia—the specialised cells which bear the spores. The dark portions (*a*) and (*b*) were covered with a thick layer of spores which would number countless millions, and so loosely heaped together that the slightest draught contaminated the atmosphere with thousands of spores. These spores measure only one five-thousandth of an inch in diameter, and their microscopical appearance is shown in Fig. 8, photographed at a high magnification. The region (*c*) is a younger growth in an active state of spore formation, and when found was exuding tear-like drops of water, a characteristic reflected in the specific name *lacrymans*.

Apart from the dangerous infective character of these fruiting bodies of dry-rot, they are unclean excrescences to have about business premises, as the old growths harbour many kinds of insect pests, mites, etc., and also accumulate much dust on their surfaces.

Having described the fruiting stage, our steps may be retraced, taking the spore as a starting point for the purpose of considering the action of the fungus on wood. Under suitable conditions of temperature, moisture and food supply the spore produces a germ-tube which develops into a hypha. The latter, branching and re-branching, soon becomes a network of threads, termed the mycelium, which, at a thousand points begins to penetrate the walls of the tubes and cells composing the wood upon which it has germinated. The substance of the woody tube-walls consists basically of cellulose, infiltrated with a substance or mixture of substances known as lignin. It is the lignin that binds and keeps the woody cell rigidly intact, and although it requires powerful oxidising chemicals for its decomposition in the laboratory it is

readily acted upon by substances secreted by the fungus. Each of the minute fungal threads possesses the property of secreting peculiar substances called enzymes, which have the power of converting insoluble substances into soluble ones, and thus rendering them available to the fungus for tissue production and energy. Penetration of a cell-wall by hyphæ is followed by the dissolution of the wall, leaving a cellulose network which is much more resistant to the action of fungi, but which, having no rigid framework to hold it together, crumbles to powder on the application of the slightest pressure. Fig. 9 shows a piece of wood from which the lignin has been removed by the action of fungi, and it consists of a collection of pure cellulose ribbons, lighter than a feather, and which has necessitated the most delicate handling.

Woody tissues also contain many other substances such as resins, gums, oils, glucosides, etc., and different fungi secrete the different enzymes necessary to effect their solubilisation. In some cases a single species is capable of bringing about changes in all these substances. *Lenzites saeppiaria*, a wood-destroying fungus previously mentioned, has recently been investigated in this connection by Zeller,⁴ who has obtained remarkable evidence of its enzyme-producing capacity. It secretes maltase, invertase and raffinase, which enable it to take immediate advantage of sugars present in an unassimilable form; diastase which converts starch into sugars; emulsin which enables it to decompose glucosides like amygdulin, salicin, etc.; ligninase, cellulase, hemicellulase and pectinase, whereby it can enter, destroy and assimilate the products of cell-walls and the substances composing them; as well as nuclease, proteinases, and oxidising and reducing enzymes. There is no kind of vegetable tissue that would not be amenable to the action of some of these enzymes, and to a large extent the fungus of dry-rot is known to be similarly equipped.

The fungus is thus able to penetrate the soundest timber and bring about those changes mentioned in our definition of decay. Fig. 10 shows a mycelium, A, that has developed on the end of a log from which the fragment of wood shown has been taken. It is growing on the surface representing a transverse section of

⁴ ZELLER, S. M., "Studies in the Physiology of the Fungi. II. *Lenzites saeppiaria* Fr., with special reference to enzyme action."—Ann. Missouri Bot. Garden, 3, 1916.

the wood and it has penetrated down into the microscopic tubes as well as between the layers. It will be noticed that a number of cracks have appeared which radiate from the centre in the plane of the medullary rays and concentrically in the plane of the annual rings. If, now, the wood is separated at these cracks,

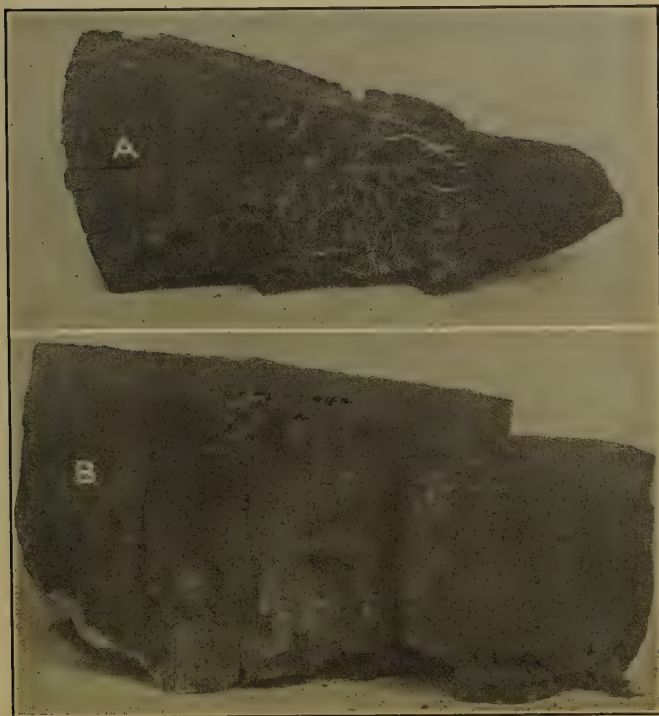


FIG. 10.

which requires little effort, radial and tangential sections will be obtained which each show ramifications of the mycelium in every direction, B. These figures are natural size, and a microscopical examination of a section much less than a thousandth of an inch

thick would show exactly the same state of affairs on the corresponding smaller scale.

Fig. 11 represents a piece of pine received from a brewery in the South of England. There are some features that lead us to suspect some fungus other than dry-rot, possibly *Lenzites saepeiaria*, but no fruit bodies have been seen by which it can be determined. In addition to the mycelium having penetrated the individual wood vessels, it has formed felty, ribbon-like growths between the rows of tubes, which, by their pressure, have had the effect of forcing whole layers of cells apart, and thus weakening the timber to such an extent that it can be picked to pieces with the fingers quite easily.

Space will not permit a discussion of the influences of temperature humidity, oxygen tension and other physical factors in the destruction of wood by dry-rot. Unfortunately in mills, breweries and other works, both temperature and moisture



FIG. 11.

conditions are usually just those required by the fungus. When the effects of dry-rot have reached the stage shown by the wood in the last figure it is obviously too late to take any steps other than replacement by sound timber. Even then, there is always the danger that infection may have spread to other structures, although there may be no outward manifestation of it, and careful inspection for signs of dry-rot should be made. In any case all the timber in a building once infected with dry-rot should be treated with some fungicide which will kill the fungus and at the same time render the wood unsuitable for any future development.

Many preparations and methods of treatment have been recommended from time to time and the impregnation of timber with a view to its preservation has been practised for more than 100 years. The substances recommended range from crude creosote to copper sulphate and other inorganic salts.

The dry-rot preventative and general fungicide developed in our own laboratories, known as "SPOROCIDE," is based on the composition of a pre-war continental preparation which consisted of potassium ortho-dinitrocresylate. "SPOROCIDE" contains corresponding sodium salts of nitro-cresylates with an important modification due to the incorporation of fluorides, the antiseptic and disinfectant values of which are well known.

Quoting from our pamphlet on this subject,⁵ its value as a germ-destroying substance was first tested in the following manner: "The walls, ceilings and woodwork of some notoriously 'smelly' cellars, after thoroughly cleaning in the usual way, were inoculated with various mould fungi, and after allowing a sufficient period for development to take place, large patches were treated with 'SPOROCIDE' and a number of preparations claiming similar properties. It was found that 'SPOROCIDE' applied in accordance with our instructions greatly exceeded in efficiency all the other products experimented with. A three-per-cent. solution immediately destroyed all colonies of mould. When a five-per-cent. solution was employed no mould fungi could be found on the surface exposed for over one year under conditions most favourable for the propagation of micro-organisms."

⁵ "Sporocide Disinfectants."—(A copy of this pamphlet may be had upon application.)

Since that paragraph was written further improvements in the efficacy of "SPOROCIDE" have been effected along lines indicated by a fuller knowledge of the physiology of the fungi. It might be argued that it does not follow from results obtained with "SPOROCIDE" on the mould fungi that it will be inimical to the fungus of dry-rot, but in addition to our practical experience that it is, there are good reasons why it should be. Fungi have been found to be extraordinarily susceptible to changes in the reaction of the medium in or on which they may be growing. As the non-technical reader will have gathered from other papers which have been published in the *Bulletin* from time to time, there are means of measuring and expressing extremely minute differences in reaction. These sensitive methods have been applied to the study of media used for the growth of fungi and the optimum and inhibiting conditions, with reference to reaction, are accurately known for a number of species.

The limits of acidity and alkalinity within which some of the moulds will grow are much wider than those found to apply in the case of the higher wood-destroying fungi. Species of *Penicillium* will develop on media between wide limits while a species of *Fusarium* will grow in a medium with a reaction on the alkaline side of neutrality much greater than that found for any other species so far studied. The reaction of "SPOROCIDE" has been adjusted in accordance with this knowledge, and it is a physiological impossibility for a fungus to develop on any substratum impregnated with "SPOROCIDE." The blue mould, *Penicillium expansum*, which is one of the most ubiquitous species in the world and one that will grow on the most diverse substances, ranging from jam to old boots, is used as a control organism in the standardization of "SPOROCIDE." A medium containing 0.008 per cent. "SPOROCIDE," but in all other respects perfectly adapted for the nutrition of *P. expansum*, retards the development of this fungus. At a concentration of 0.015 per cent. the mycelium grows but does not fructify, and at 0.02 per cent. "SPOROCIDE" entirely prevents the germination of conidia, no growth whatever being found in the tube heavily inoculated with the fungus.

F. A. M.

The Suppression of Insect Pests and Fungoid Diseases

3. THE FUMIGATION OF COMMERCIAL GLASSHOUSES IN THE GROWING SEASON

IN the previous issue of the *Bulletin* (No. 8) reference has been made to the fumigation and disinfection of glasshouses. This, of course, only refers to the treatment when completely empty and in the dormant period. It is proposed in this article to outline the various methods at present practised for controlling both pests and diseases under glass.

It is now generally acknowledged that Fumigation is the best, surest and most economical way of treating a glasshouse for pests and is to be preferred to either Dry Dusting or Spraying for the following reasons:—

1. It is scientifically sound in principle because it involves the use of a toxic vapour or gas which can permeate all parts of the house by reason of the ease with which it mixes with air. This process of mixing gases is termed diffusion, and was first elucidated by the chemist Graham. He found that lighter gases diffuse or will mix more rapidly than the heavier ones, and stated a law which applies to all gases—namely, “That the relative rates of diffusion of gases are inversely proportional to the square roots of their densities.” This may require a more explanatory statement.

Hydrogen gas has a density of 1. Air, as compared with hydrogen, is 14.4 times heavier and is said to have a density of 14.4. Carbon dioxide or soda-water gas has a density of 22, so that one would expect hydrogen and air to mix more readily and rapidly than air and carbon dioxide—this does agree with what is found in actual practice. The rate of mixing is also influenced by temperature and proceeds at a greater rate with increased temperature. Convection currents are also set up which assist the process of diffusion.

2. Fumigation therefore is quicker and more effective, providing of course always that the gas has a sufficiently high insecticidal value and is present in sufficient concentration to be lethal to the insects compatible with the length of time the house is closed for fumigation.

Dry Dusting is the nearest approach to fumigating that is possible. It consists in impregnating some inert carrying material with the insecticide, which must be volatile.

The efficacy of dusting largely depends upon the fineness of division of the carrying material, its density or floating capacity, and the toxicity of the insecticide and the means of distribution. Therefore, fumigation wherever possible is to be preferred to dusting.

Unfortunately the number of volatile fungicides which can be used for fumigating are comparatively few. It is, on the other hand, questionable whether a volatile fungicide would control or prevent the entrance of the hyphæ or germ tubes through the epidermis of the leaf or its stomata.

The writer is of the opinion that the efficiency of a fungicide largely depends upon (1) its solubility in water and (2) its intimate contact with the spore or hypha of the fungus, in order that the active principle may penetrate by diffusion the cell wall and cause coagulation of the protoplasmic fluid inside the spore or hypha. Dry Dusting in the presence of moisture or wet spraying must be

resorted to in such cases. For many reasons the former method is to be preferred to the latter for reasons which will be discussed later.

The fumigation of a mixed house is always attended by a certain amount of risk owing to the greater or lesser susceptibility of different varieties of plants to chemical absorption. It is important as a first consideration to ascertain the effect of a fumigant upon a representative collection of plants on a small scale before proceeding to fumigate on a large scale, otherwise disastrous results may follow, because it is well known that certain volatile fumigants, whilst quite safe to use upon such plants as cucumbers and tomatoes, are very dangerous to use upon chrysanthemums and cinerarias. At present it is impossible to say why a sappy plant such as a cucumber should survive whereas a half-hardy plant like a chrysanthemum will succumb to the same treatment. No doubt in the light of modern research an explanation before long will be forthcoming.

The next point for consideration is the selection of the Fumigant. Certain compounds are particularly toxic to one species of pest whilst they may have little, if any, effect upon others. On the other hand, there are insecticides which can be used upon a large variety of insects with success simply by altering the concentration and mode of application. It is therefore important to use the right fumigant. In any system of fumigation there are certain essential conditions which must always be observed. These are enumerated below:—

1. Correct estimation of cubical capacity: for all practical purposes the length multiplied by the breadth multiplied by half the average height will give a near estimate.
2. The maintenance of definite limits of temperature and in some cases humidity.
3. The desirability of fumigating during the hours of darkness when plant inspiration and respiration is at a minimum owing to the partial closing of the stomata.

4. The application of the fumigant at such a concentration that it will be lethal to the insect without producing any deleterious effects on the plant.
5. The length of time the fumigant should be allowed to act is most important. In many cases a low concentration fumigation extending over a number of hours is to be preferred to one of shorter duration at a higher concentration. In other cases the reverse conditions have given better results. In determining this point, plant resistance, stamina and vitality, susceptibility to chemical absorption must all be taken into careful consideration.

THE CONTROL OF WHITE FLY ON TOMATOES.

According to Dr. Lloyd (*Annals of Applied Biology*, Vol. 9, No. 1, April, 1922), this pest is able to withstand a mild winter out of doors.

It breeds extensively outside during the summer months on a variety of plants, particularly the stinging nettle. It favours warm conditions and is responsible for considerable damage under glass, by excreting a sticky honeydew which covers both fruit and foliage and necessitates thorough wiping of the former before marketing. Two methods are recommended for fumigating against this pernicious pest, namely:—

1. Cyanide, and
2. Volatile Chlorinated Hydrocarbons.

The former method has been in use for some years, but it is now being rapidly replaced by the safer and latter materials.

In Cyaniding there are, in addition to the general conditions previously mentioned, a number of special conditions which must be rigidly adhered to in order to obviate damage to plants and to secure success with the treatment.

Temperature.—Generally speaking, a house should be cyanided at as low a temperature as possible.

Time of day.—It is essential to fumigate only at dusk when the sun has set, otherwise disastrous results will be obtained. The cyanide gas appears to be less toxic to insect life in the presence of sunlight.

Fumigations of long duration using lower concentrations of the gas are better than those of short duration at the greater concentrations.

Effect upon plants.—The same author (Lloyd) considers that the Tomato plant is particularly susceptible to damage by cyanide gas, hard-growing wiry plants resisting the treatment better than soft sappy ones. Fully-grown leaves appear to be less liable to damage than the younger ones. Check upon plant growth does occur as a result of cyaniding and this will depend to some extent upon local conditions.

It has also been found that it is necessary to dry off the roots before fumigating so that the plants are not in a very turgid condition, otherwise severe damage results.

FUMIGATING WITH VOLATILE HYDROCARBONS.

Amongst the various materials used in Lloyd's experiments was Tetrachlorethane, a volatile fluid which proved particularly good in killing the fly without damaging that plant at the rate of 5 to 10 fluid ozs. per 1,000 cu. ft. of space. At that time (1920) it could not be made a commercial proposition owing to its high cost and the high concentration required.

Continuing work on the lines of Lloyd, we were able, after a considerable amount of experimenting, to compound a volatile fumigant which could be used at 2-4 fluid ozs. per 1,000 cu. ft., depending upon temperature, tightness of the house and the degree of infestation of the pest.

This material has been extensively used by growers in this country and the Channel Islands with considerable success.

Its great advantage lies in the simplicity of its application, requiring no apparatus, acids or special conditions as with Cyanide, and without the disadvantage of a check on plant growth or the

necessity for drying of the plant roots prior to fumigating. Although, perhaps, slightly more costly, this method possesses many distinct advantages over the older and almost obsolete method of Cyaniding and without the recognised disadvantages of cyanide, being also non-poisonous.

The only essential condition to be observed when applying is to have the house temperature as near 70°F. as possible and not below 60°F., otherwise the product does not evaporate at a great enough rate, and the fumes become dispersed before they have concentrated in sufficient quantity to become deadly to the pest.

In addition to being suitable for Tomatoes, this fumigant may be used upon a large variety of plants. (For information on this point, enquiries should be addressed to our Testing Station.)

CONTROL OF GREEN FLY UNDER GLASS.

Nicotine in one form or another has been found to be the most effective insecticide for use under glass in controlling Aphis.

It can be applied as a Dry Dust as previously indicated, and in such a form has given excellent results. Fumigation with Nicotine requires a special vaporiser owing to the high boiling point of the liquid, its rate of evaporation in an ordinary hot-house being much too slow to be effective.

It has been put up in another form by impregnating brown paper with the insecticide in such a quantity that the paper when ignited gradually smoulders, causing the volatilisation of the insecticide.

The use of liquid spray fluids in a Tomato house is to be deprecated, because under certain weather conditions the introduction of moisture in the form of an insecticide into the house, in some districts, may cause the appearance of *Cladosporium fulvum*.

The danger is not so real if the insecticide is fungicidal at the same time.

In a subsequent issue the control of other pests and diseases under glass will be dealt with.

T. P.

Nicotine Petroleum Emulsion

A COMBINED INSECTICIDE

IN the early days of insecticides much attention was devoted to the use of Kerosene or Petroleum as an insecticide.

With the progress of science many new products have come into use and many old ones have been discarded, yet, in spite of this, Petroleum still ranks as a first-class insecticide.

In this country it fell into disfavour largely due to the fact that it was not properly emulsified and so caused a certain amount of scorching of foliage.

One great advantage with Petroleum is that it can be successfully compounded with other types of insecticides to form compatible mixtures which undoubtedly have a greater insecticidal value than the single ingredient. Such a mixture may be termed a combined insecticide as distinguished from a combined wash, which is usually understood to mean a combined insecticide and fungicide.

In the former there is a combination of ingredients, in the latter a combination of ultimate properties, and it will be obvious if the combination of ingredients be carefully arranged, a greater variety of types of pests can be controlled by the one process. Thus, by the use of a combined insecticide, economy is effected in addition to the insurance of effective results.

It might be well at this point to outline briefly the various types of insecticides which could be compounded together as above described. Generally speaking, they may be classified as

1. Stomach poisons.
2. Contact washes.
3. Asphyxiants and Fumigants.
4. Narcotics.
5. Deterrents.

Class 1 is usually reserved for leaf-eating and biting insects.

Classes 2, 3 and 4 may be successfully used in cases of young soft-bodied caterpillars, etc., in addition to Class 1.

Classes 2, 3, 4 and 5 are all used against leaf-sucking insects.

Examples of the above classes are as follows:—

Class 1.—Arsenate of Lead, which eaten with the foliage rapidly poisons the insect owing to internal absorption.

Class 2.—Coal Tar Oils of high boiling point for winter use only. Petroleum Oils, emulsified and used with a suitable “spreader,” cause an irritating action upon the skin (if not too tough and hardened). At the same time, owing to capillary attraction and reduced surface tension of the wash as a result of the presence of the spreader, it gradually penetrates the spiracles, and entering the trachea, causing asphyxiation.

Class 3.—Soap or other spreaders which act in the same manner as those in the latter part of Class 2. Various toxic gases, such as carbon bisulphide, naphthalene vapour, pyridine, sulphur gas, cyanide gas.

Class 4.—Various members of the group of natural products which include Nicotine, and which, on coming into contact with the skin of a soft-bodied insect, are rapidly absorbed, causing paralysis of the nervous system.

Class 5.—Deterrents.—Simply the application of some material to the surface of the plant which will be distasteful to the insect

and prevent it from inserting the proboscis into the epidermis of the leaf. Such substances as bitter aloes and quassia are used for this purpose.

It will be readily seen that by the judicious mixing of representatives of the various classes it is possible to produce a wash or insecticide which enables one to attack a pest from a number of points of vantage.

At the same time it must be borne in mind that such a mixture must be compatible, that is to say, the various ingredients must not react with one another chemically and produce a compound which may be innocuous to the insect but may damage the plant.

Soap is usually one of the constituents of a wash, the object of its incorporation in a spray fluid being to assist the spreading over the foliage and pest.

In the United States much work has been done on the various classes of soap, and many authorities favour the use of a potash fish-oil soap, as, in addition to its action as a spreader, it possesses insecticidal properties of both a chemical and physical nature.

Experimental work at our own Station carried out on this particular type of soap has shown that potash resin fish-oil soaps generally are to be recommended, as they possess, in addition to excellent spreading properties, a fairly high insecticidal value. The results obtained with such a soap have been so satisfactory that we now incorporate the necessary proportions with other ingredients in the various washes we recommend for special purposes.

On this account it has been possible to prepare a combined insecticide which embraces the classes 2, 3 and 4 types of insecticides in the form of Nicotine 2 per cent., Petroleum oils 50 per cent. (special specification), and which is now known as Nicotine Petroleum Emulsion or N.P.E. This spray has had an

extensive use amongst commercial growers for controlling Aphids of different species, Capsid Bugs, Apple and Pear Sucker, young Scale insects, Red Spider on cucumbers and tomatoes, and Woolly Aphis on various plants.

This emulsion is in reality a colloidal emulsion or solution. With an ordinary oil emulsion (non-colloidal), the particles are in a state of fine suspension and later as fine globules which are easily discernible under an ordinary high-power microscope. In a colloidal solution the particles are in a much finer state of sub-division and they cannot be seen as individuals with ordinary means. They can, however, be observed with the aid of a special instrument called an ultra microscope.

A colloidal solution when in a diluted condition after a time begins to change and there is a gradual transition in the size of the particles from an invisible to a visible form.

It is well known that when matter changes from one phase to another a certain amount of energy is liberated, and it is very probable that this energy when set free and when associated with the transitional stages above mentioned in connection with N.P.E. greatly increases the insecticidal value of such.

These theoretical considerations are borne out in actual practical tests carried out with N.P.E. In any Petroleum Emulsion with high concentration of the active ingredients there is always a difficulty in obtaining good penetration and spreading over the foliage and pests. This is due to its high surface tension and the degree of hardness of the water used for diluting the spray.

In any case a very hard water is undesirable, as there is a tendency to cause de-emulsification of the oils. Also the presence of fair quantities of chloride in the water produces a salting-out effect on the soap with production of a thick scum which may cause considerable spray-nozzle trouble as well as a de-emulsifying effect on the wash. The use of a spreader such as calcium caseinate greatly assists the spreading, and, at the same time, owing to the

reduction of surface tension, causes the insecticide to come into more intimate contact with the pest, thus securing its maximum effect.

It has been found, for instance, that N.P.E. of the specification above described can be used at 1-160 dilution in the open for controlling Aphids, Psylla and Capsid Bug if used in conjunction with 0.2 per cent. calcium caseinate. Whereas, when used by itself, it is necessary to use the material at nearly double the concentration.

By using Nicotine Sulphate Petroleum Emulsion (that is, by replacing the pure nicotine with an equivalent amount in combination as a sulphate) in conjunction with calcium caseinate Woolly Aphis has been successfully controlled when the pressure was sufficient to permit of the use of a coarse nozzle to ensure penetration of the woolly protective covering of the Aphid.

The same specific has been found most effective under glass for controlling Red Spider on cucumbers and tomatoes at a dilution of 1-200, by spraying in the evening and, during the hot weather, washing the plants with clear water on the following morning.

A repetition of the treatment is of course necessary to ensure complete control.

Providing the spreader has been used with N.P.E., no damage has been noticed in the form of "spotting," either under glass or in the open.

In N.P.E. we have a compound which embraces the various classes of insecticides previously indicated, and on this account, and in view of its colloidal state, it should find extensive use among commercial growers generally.

T. P.

NOTES

THE GREENHOUSE GRASSHOPPER.

We have received a communication from Prof. Dr. Richard Ebner, of Vienna, to whose paper on this insect reference was made in our article in the last issue of the *Bulletin*, in which he kindly informs us that *Tachycines asynamorus* is a native of Eastern Asia. We are pleased to hear that Prof. Ebner hopes to deal further with this pest in a future publication.

LECTURES ON HORTICULTURE.

On February 26th and 27th two lectures were delivered in Guernsey by the director of our Agri-Horticultural Testing Station, Mortlake. At St. Peter's Parish Church Schools and at Elizabeth College Hall, Mr. Parker spoke on "Scientific Methods of Treating Soil" and "Greenhouse Sanitation" respectively. These lectures were fully reported in the *Star*, Guernsey, February 28th, and the gist of the matter discussed is embodied in articles contained in this and in previous issues of the *Bulletin*.

Micro-Organisms

in the

Leather Industries

4.—THE GENUS ASPERGILLUS

THE moulds included under the generic name, *Aspergillus*, like those belonging to the allied genus, *Penicillium*, are among the most widely-distributed fungi in the world. Growths of different species of *Aspergillus* may be white, blue, green, yellow, brown or black, and a species of the latter colour was one of the first organisms ever associated with leather. The structure of a member of this genus is characterised by a colourless, spreading, septate mycelium, which gives rise to fertile hyphæ varying in length from 0.2 mm. to about 1 cm., according to the species, although individual plants of the same species may show considerable variation within these limits. The tips of the fertile hyphæ are inflated, the vesiculose portion of each forming a receptacle upon which are developed numerous simple or branched sterigmata. In the latter case the basal sterigmata may be termed the *primary*, and the usually shorter branches, *secondary* sterigmata. From the apices of the sterigmata arise chains of conidia, as in the case of *Penicillium*. The essential features of the conidial stage of *Aspergillus* are shown in the diagram, Fig. 12.

It will be remembered, or it may be ascertained on reference to our systematic arrangement of the fungi, *Bulletin* No. 3, p. 78, that *Aspergillus* is placed among the Fungi Imperfecti, thus indicating that it represents only a stage in the life-history of some

other and more highly developed fungus. The Fungi Imperfecti are usually asexual forms of Ascomycetes, although the relationships of the great majority of the species still await discovery. In the case of *Aspergillus*, however, the perfect stages of some of the species are well known, and the mycological purist might therefore be justified in ignoring the existence of the genus in the Fungi Imperfecti by regarding it only as belonging to the *Aspergillaceæ*,

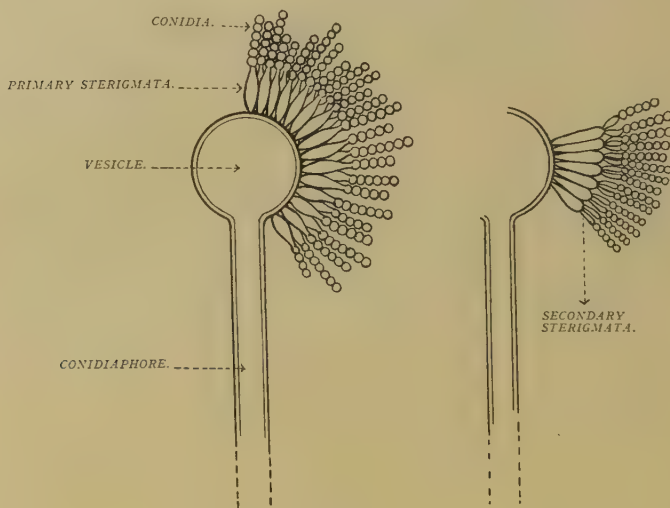


FIG. 12.

a family of the truly Ascomycetous fungi. Identification is based upon the conidial stage of the fungus, and although further reference to the perfect stage will be made when discussing *Aspergillus glaucus*, the genus will here be retained in the Fungi Imperfecti on technological grounds.

The fact that the sterigmata may show a secondary as well as a primary structure has been responsible for a great deal of confusion in the literature relating to the species, and whilst it is to be hoped that confusion on this score came to an end in 1916, on the publication of a paper by Charles Thom and James N.

Currie,¹ the general difficulties encountered in accurately naming species are very similar to those mentioned in our earlier notes on the species of *Penicillium*.

The confusion arising from the structure of the sterigma has been responsible for at least two names for one species in the literature of leather technology, to say nothing of at least eight other names given to the same species, which occur elsewhere, and about which the leather worker need not trouble. The species referred to is *Aspergillus niger* Van Tieghem, a black mould known equally well as *Sterigmatocystis niger* Cramer. The existence of these two names is explained by the fact that the genus *Aspergillus* was originally founded on a plant which possessed simple sterigmata, and when a similar fungus possessing branching sterigmata was discovered, the name *Sterigmatocystis* was applied, and it became customary to separate the two genera as follows:—

- (a) Sterigmata of apical vesicle simple or nearly so
= *Aspergillus*.
- (b) Sterigmata of apical vesicle verticillately branched
= *Sterigmatocystis*.

It has since been found that species of either the one or the other genus when grown in pure culture from a single conidium would sometimes exhibit branched, and sometimes simple sterigmata, whilst it might also show both types. It is therefore obvious that the occurrence of secondary sterigmata is not even a constant specific character, and it can possess no significance from a generic point of view. This fact was recognised by Wehmer in 1901, and he emphasizes it in his contribution to Lafar's *Technical Mycology*,² although he there adheres to the name *Sterigmatocystis* in his systematic treatment of the species of *Aspergillus*.

Further confusion, especially in regard to the various black or fuscous species, has operated in two ways. In one direction by the creation of numerous species on grounds merely of difference in substratum, e.g., *Sterigmatocystis ficuum* found on figs, *S. anta-*

¹ "Aspergillus Niger Group": *Journ. Agric. Research*, U.S. Dept. of Agric., Vol. VII, 1916, p. 1.

² Eng. Ed., 1910, Vol. II, Part II, p. 296 et seq.

caustica, discovered as a pathogen in the human ear, etc., all of which have been insufficiently described, and which, in all probability, are merely forms of *Aspergillus niger*. Whilst there are undoubtedly a large number of well-differentiated black species of *Aspergillus*, chemists and physiologists, with few exceptions, have gone to the other extreme and have designated all the black species with which they have worked, *Aspergillus (Sterigmatocystis) niger*.

A striking illustration of the futility of carrying out physiological work on organisms not carefully diagnosed is brought out in a Table given by Thom and Currie in the paper already cited. The property of producing relatively large amounts of oxalic acid from suitable carbohydrates has long been associated with *Aspergillus niger*, and the authors made experiments with a view to demonstrating the comparative oxalic acid production by strains of *Aspergillus niger* and other black, nearly related organisms, grown in flasks of 50 cc. each of Czapeks solution containing 5 per cent. of sugar. These were grown for periods of 7, 10 and 18 days in each case. At the end of each period the acidity and oxalate radical were determined and expressed in cubic centimetres of N/10 sodium hydroxide required to neutralize 50 cc. of the medium. The following particulars of six species out of twenty tested will indicate the extent of variability in acid production :—

NAME OF SOURCE.	7 DAYS.		10 DAYS.		14 DAYS.		18 DAYS.	
	Acidity.	Oxalate.	Acidity.	Oxalate.	Acidity.	Oxalate.	Acidity.	Oxalate.
<i>A. ficum</i> (?) ...	116·17	116·21	142·85	155·82	167·90	182·90	153·21	170·01
<i>A. niger</i> (Amsterdam)	62·30	67·82	80·52	94·72	79·54	94·35	71·12	87·05
Pittsburgh ...	103·74	48·57	97·96	79·95	89·92	85·56	61·14	60·34
Soil, England...	29·78	30·64	58·78	74·73	29·58	45·59	22·92	39·42
<i>A. fuscus</i> ...	85·23	81·08	26·64	40·73	19·39	33·07	12·34	28·02
<i>A. carbonarius</i> ...	31·54	14·57	5·58	18·31	6·66	21·33	3·53	13·85

Two other forms which could be readily distinguished from the typical black group were included in the test, and these gave the following figures:—

NAME OF SOURCE.	7 DAYS.		10 DAYS.		14 DAYS.		18 DAYS.	
	Acidity.	Oxalate.	Acidity.	Oxalate.	Acidity.	Oxalate.	Acidity.	Oxalate.
<i>A. ochraceus</i>	1·57	7·10	1·47	13·08	0·78	14·70	0·39	11·58
<i>A. violaceo-fuscus</i> (?)	17·73	9·15	12·73	18·87	9·01	17·56	3·13	16·44

Thom and Currie conclude that the wide variations in oxalic acid production found in their experiments indicate either a group of heterogeneous ancestry or a series of races of a single ancestry, which show great variation in the ability to produce a particular reaction. They point out that if such variation can be correlated with morphological characters, it is a valuable accessory in the identification of species. If cultures exhibit only quantitative differences in the reactions selected for study, such differences may be exceedingly important economically without justifying the description of separate species.

Arising out of further cultural and physiological studies of the black forms of *Aspergillus*, the authors reach conclusions, of which two are important from our present point of view. The first is that *Aspergillus niger* and a number of allied organisms form a group that may be held to consist of a series of forms, some of which seem to be connected so closely by intergrading forms as to make separation difficult, if not impossible. For this series of organisms the name "Aspergillus Niger Group" is proposed. The other conclusion, already mentioned, is that the examination of many thousands of specimens does not justify the use of the separate name *Sterigmatocystis*.

The latter view does not necessarily mean that the presence of secondary sterigmata is altogether without importance in assisting diagnosis. On the contrary, the regularity with which differences in this respect are exhibited is made the basis of a

useful key to the black species, so long as it is fully appreciated that the organisms determined by it merely fall within the *Aspergillus niger* Group.

The following are the sub-divisions of this Group :—

1. Forms with simple sterigmata up to 20μ in length.
A. nanus MONTAGNE.
2. Forms with both primary and secondary sterigmata ;
primary $20-30\mu$ in length.
 - (a) Black or brown.
A. niger VAN TIEGHEM.
 - (b) Forms differing from *A. niger* only in colour.
A. cinnamomeus SCHIEMANN.
A. Schiemanii (SCHIEM.) THOM.
3. Forms with both primary and secondary sterigmata ;
primary about 50μ in length.
A. phoenicis (CORDA) PATOUILLARD and DELACROIX.
4. Forms with both primary and secondary sterigmata ;
primary up to 120μ in length.
A. pulverulentus MCALPINE.
5. Forms with both primary and secondary sterigmata ;
primary up to 120μ in length, spores double size.
A. carbonarius (BAIN.) THOM.

The only organism belonging to this group so far found in connection with leather industries is *Aspergillus niger*.

(TO BE CONTINUED.)

F. A. M.

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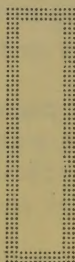
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